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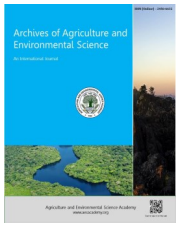

ORIGINAL RESEARCH ARTICLE

Toxicity of non-selective herbicide-Paraeforce® to Periwinkle snail *Tympanotonus fuscatus var radula*

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ABSTRACT

Periwinkle snails *Tympanotonus fuscatus var radula,* an important shellfish in the Niger Delta ecological zone are on the decline and they have not been considered in environmental risk assessment due to the lack of standardized protocols. The gastropod is an abundant species and widely distributed in the aquatic Niger Delta environments. In this study *T. fuscatus var radula* was exposed to ParaeForce(e), a solution of paraquat dichloride, a non-selective commonly used organochlorine herbicide due to it sensitivity to chemical compounds, as pollution indicators. The effect of lethal and sub lethal exposure of ParaeForce® was assessed using the Organization for Economic Development and Cooperation (OECD) #218 protocol to determine the toxicity and safe limit concentrations. The lethal exposure gave an average LC_{50} of 0.665 mg/kg with a safe limit of 0.0665 mg/kg. The sub lethal test showed that the exposed organisms reduced significantly in body mass at levels of $P < 0.05$ when compared to the control experiment. Mean percent growth rate relative to the control reduced from 100% to 47% while the highest growth inhibition of 53% was observed in the highest concentration. Thus, the release of the test chemical and other similar herbicides into the environment may cause alteration and loss of body mass in periwinkle molluscs due to their toxic potentials. It is important to exercise caution in the application of this herbicide especially in the aquatic environment for weed control.

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INTRODUCTION

The use of synthetic chemical pesticides in modern agricultural practices is well recognised as a cost-effective method of controlling or preventing agricultural pests (Liu *et al.,* 2010; Narita *et al.,* 2014). Chemical pesticides consist of an active ingredient, the actual poison, and a variety of additives, which improve the efficacy of their application and action (MacFarlane *et al.,* 2013). They are classified or grouped according to the target organisms, chemical structure of the compound (Benavides *et al.,* 2006; Alavanja *et al.,* 2004), or type of health hazard involved (WHO, 2010). The release of pesticides from agricultural fields and the resulting contamination of the environment may pose both ecological and human health risks (Capri and Karpouzas, 2007). Given their globally application and the fact that they are designed to harm biota, there is a high potential for adverse environmental effects especially on non-target communities (Van der Werf, 1996). Pesticides when improperly selected and managed, they can pollute water resources with carcinogens and other toxic substances that can affect humans and many forms of fish and wildlife thereby posing serious

threat to public health and biodiversity loss with negative impacts in the food chain.

Herbicides are the most widely used class of pesticides in the world to suppress or kill unwanted vegetation (weeds) (Mensah *et al.,* 2014). They are only one of the many types of pesticides that include insecticides, fungicides, rodenticides and nematocides (Jurado *et al.,* 2011). Every year about six thousands of tons pesticides are deliberately introduced into the environment in order to prevent loss of agricultural crops (Lovecka *et al.,* 2015). Even at low concentrations, pesticides still could elicit deleterious effects, such as cytogenetic damage, physiological effects, and even death, to exposed non-target species (Dinis-Oliveira *et al.,* 2008)

Paraeforce (e) a paraquat dichloride is a water soluble herbicide registered to control weeds and grasses in many agricultural and non-agricultural areas (USEPA, 1997). It is among the leading products used in Nigeria to control both aquatic and terrestrial weeds and invading alien plant species. In agriculture, it is use in pre-plant or pre-emergence on vegetables, grains, cotton, grasses, sugar cane, peanuts, potatoes, and tree plantation areas; post emergence around fruit crops, vegetables, trees, vines, grains, soybeans, and sugar-cane; during the dormant season on clover and other legumes. It is also used as a defoliant and desiccant to aid in the harvesting of cotton, beans, soybeans, potatoes, sunflowers, and sugarcane (Gwathway and Craig, 2007) and as a post-harvest desiccant on staked tomatoes. Paraeforce is also used in non-agricultural areas such as roadsides, airports, around commercial buildings, drains, irrigation ditches, and waterways. Paraquat is registered and sold under different trade names, such as Gramoxone, Crisquat, Dextrone X and Esgram. Paraquat is on PAN International's Dirty Dozen (1985) and Highly Hazardous Pesticides (2009) lists for global phase-out. It has been banned in 32 countries (including the countries of the European Union), mainly for health reasons and environment persistence since 2007. However, there is strong industry resistance to including paraquat in the Rotterdam Convention on Prior Informed Consent and it remains outside the PIC list.

Besides the benefits derived from the use of paraquat herbicide in forestry and agriculture, they also have serious negative impacts on human health via contamination of food and waters. Over the last decades in Sub-Sahara Africa, cases of acute human poisoning account for significant morbidity and mortality. In Nigeria, there is sufficient evidence that suggest large scale and trends of problems caused by pesticides. In 2015, the World Health Organisation (WHO) reported pesticide poisoning that led to 18 mysterious deaths in south-western Nigeria. Also, in Cross River State 112 people were hospitalised from food poisoning of which two children died. Again in Gombe state, over 120 students were hospitalised after consuming a meal of beans suspected to have been preserved with poisonous chemicals. The laboratory reports on these incidences revealed high levels of organophostate, carbamates, fenithrothion and chloropyrifos toxic pesticides in food substances.

Today there is a growing public awareness of the increase herbicides use and their adverse effects on aquatic ecosystems (Pérez *et al.,* 2011). In Nigeria, the widespread use of pesticides over the years has resulted in the accumulation of pesticide residues in the environment with negative effects on biota and ecosystem functions. For example, Periwinkles a gastropod snails species, have been on the decline in Nigeria due to female masculization a condition known as imposex as a result of endocrine disrupting chemical in aquatic environment (Ogbomida and Ezemonye, 2013). Herbicides residues in aquatic environment may sink into the sediment due to their repeated use and high solubility. Herbicides may reach water bodies indirectly through agricultural runoff, spray drift and leaching or direct overhead spray to control noxious aquatic weeds (Marin-Morales *et al.,* 2013). Once in the aquatic ecosystems, herbicides may reduce environmental quality and influence essential ecosystem functioning by reducing species diversity and community structures, modifying food chains, changing patterns of energy flow and nutrient cycling and changing the stability and resilience of ecosystem (Pérez *et al.,* 2011).

Various studies in Nigeria have reported adverse impact of pesticides on non-target animals in other to address contaminants in the aquatic environment. However, their effects on gastropod snail sensitivity are still very limited despite increased use of agrochemical products to improve crop yields. This phylum is the second largest group in the kingdom animalia and is vital to sustain many ecosystems. Molluscs are considered excellent indicators of ecosystem health in general, and because they are particularly sensitive to changes in their environment, they can act as early warning sentinels of habitat deterioration (Wells and Chatfield, 1992). Therefore, this study aimed to determine the acute and sub lethal effects of a non-selective herbicide - ParaeForce® to periwinkle (*Tympanotonus fuscatus var radula*) with a view to develop data for maintaining the quality of our water resources.

MATERIALS AND METHODS

Ethics statement: Periwinkle snails used for this procedure were collected from public natural brackish water environment in the Niger Delta Ecological region. We state clearly that no specific permission was required for the locations/activities and confirm that Periwinkle snails are not in the lists of endangered or protected species.

Test chemical: Paraeforce (e) a paraquat dichloride (1:1′-dimethyl 4:4′-dipyridylium dichloride) solution was used for the static renewal sediment toxicity bioassay. It is a non-selective contact agriculture herbicide use in the control of stubborn, annual and perennial grasses and brad-leaved weeds. Paraforce was purchased from a local agrochemical shop in Warri Delta State Nigeria. Commercial preparation of ParaeForce® containing 276 g/L organochlorine ($C_{12}H_{14}Cl_2N_2$) was used as stock solution for the bioassay. Using this working stock, various concentrations of the test herbicide was prepared for range-finding and definitive (lethal and sublethal) tests. Dimethyl sulfoxide (DMSO) was used as a solvent in each concentration treatment with a final volume-to-volume ratio of 0.05% for toxicity.

Test organisms collection and acclimatization: The test organisms Periwinkle snails *(Tympanotonus fuscatus var*

radula) of the Nigerian Niger Delta ecological zone inhabit the brackish water environments. Similar sizes (shell length 4.0 ± 0.03 cm, diameter of aperture 0.07-0.09 cm, mean weight 5.474 ± 0.04 g) were collected by handpicking into a bucket (12.6L) from Maciver jetty (water-side) at low tide, in Warri South Local Government Area (LGA) of Delta state, Nigeria. The Periwinkle snails were collected from the same site (5°00 and 6°.45 E and 5°00 and 6°30 N) to reduce variability in biotype. They were washed with surface water to remove mud, transported to the laboratory and kept in holding glass tanks (30 × 30 × 30 cm) with aerated brackish water (6L) at measured salinity. The snails were gradually acclimatized to laboratory conditions (Table 1), which were based on natural habitat conditions. The sediment from the brackish environment where the snails were collected was used for acclimatization. During the acclimatization process the gastropod snail *Tympanotonus fuscatus var radula* were fed with ground aquarium fish food according to Ducrot *et al.* (2006) for best survival and growth.

Testing of sediment: Test sediment samples were collected from the top few mm of the surface sediment layer at mid-intertidal level of selected mudflats and stored in containers made of inert materials to prevent contamination. Prior to its use, sediment were kept at room temperature (21 ± 2ºC) for a minimum of 24 hours to allow it to equilibrate to the test temperature.

Bioassay procedure: The 10-day experimental procedure for sediment toxicity bioassay for periwinkle was carried out using the Organization for Economic Cooperation and Development (OECD) #218 protocol (OECD, 2004). The periwinkle snails were acclimated in unspiked sediment for seven (7) days before the bioassay test. The experiment began with a preliminary range-finding test to establish a working concentration for the definite test. From the prepared stock solution of the test chemical using dilution water from the organisms' habitat, serial dilutions were made to obtain concentrations in the range of 100, 10 and 1 mg/L (100 mg/g, 10 mg/g and 1 mg/g). The range-finding test was conducted and terminated within 24 hours.

Acute sediment toxicity test for 10 days: The acute toxicity of the Paraeforce -spiked sediment to periwinkle snails was evaluated over a period of 10 days according to the test conditions described in Table 1. The 10-day static sediment bioassay with renewal of the overlaying uncontaminated water (filtered to 0.45 µm) was used to prepare nominal concentrations of 2.0, 1.0, 0.5, 0.25 and 0.125 mg/L resulting in 2.0, 1.0, 0.5, 0.25 and 0.125 mg/kg after been placed in 1000g of uncontaminated natural sediment with known physic-chemical parameters (grain size, porewater salinity and pH). The sediment in the container was evenly spread to form a thick layer of substrate and 2000 mL of the prepared test solution was gently added. This composition in the test vessels was then left to settle for 2 to 3 hours before the introduction of the test organisms. Before the periwinkles were introduced into the test tanks, they were placed in uncontaminated dilution water to rinse off any debris adhering to the organisms and later placed on clean sheets to void stomach content. After which, ten (20) healthy periwinkles were gently transferred into each glass vessel containing the test chemical and control. The overlaying water was gently aerated for the exposure period. The overlaying water was renewed on day 3, 6 and 10. Observations for lethality in the test vessels were carried out and records of species dead or discoloured were noted. After the test period, the number of periwinkle dead or alive was recorded. The average mortality in the bioassay i.e. the total number of dead organisms related to the total number of organisms used on day 0 was used to compute the average mortality in the bioassay at day 10. In order to prove the sensitivity of the test chemical and its effect on the species, controls with clean sediment without the test toxicant was conducted (Environment Canada, 1992) i.e. dilution water and sediment from the habitat of the test species control.

Sub lethal assessment: The concentrations used for the sub lethal bio test was obtained from the results of the LC_{50} and was serially diluted to obtain the concentrations used. Active species were cleaned and the length and weight taken at day 0 and at test termination of day 28. Periwinkle snails were then transferred to the sediment spiked with different concentrations. Replicates per treatment were prepared for the exposed concentrations of the test chemical. The control setup containing dilution water was also prepared in conjunction with the test chemical but without the toxicant. The setup was covered with net to prevent the test medium from drying for the test duration. Toxicity modifying factors considered were bioaccumulation potential with specific reference to growth inhibition.

Physiological effect: The physiological endpoint used in this assessment was growth. Mean weight of the total number of organisms used for the test was taken at initiation (day 0) and termination (day 28) of the test (Equation 1). Before weighing, the periwinkles were sorted, washed with tap water, and blotted with filter paper, and then weighed. The weight of each periwinkle determined after 7, 14, 21, and 28 days of exposure were compared with controls. The weight Growth inhibition was computed using the formula (Equation 2):

$$\text{Mean weight} = \frac{\text{Weight of Organism at day 0} + \text{Weight Organism at day 28}}{2}$$

Eqn. 1

$$\text{Growth inhibition (\%)} = \frac{(CL - TL)}{CL} \times 100$$

Eqn. 2

Where C_L is the mean periwinkle weight (g) in the control and T_L is the mean periwinkle weight (g) in the treatment.

Water and sediment chemistry: The monitoring of water and sediment quality parameters were taken at random at the start, during and termination of the test. The physico-chemical parameters of the overlying water and sediment were determined to provide relevant information on possible changes that could result in potential hazards to the biological indicators. Physico-chemical constituents determined in water include; pH, temperature, dissolved oxygen (DO), salinity and conductivity while pH and total organic carbon (TOC) in sediment.

Assessment of mortality: Percentage mortality (endpoint indicator of acute toxicity) was evaluated on day 10 of the experiment in all replicates. Physical changes (morphology)

and behavioural changes were also noted. Organisms were considered dead if there was no movement or if there is no activity after few minutes of placing the periwinkle on moist white filter paper.

Statistical analysis: The vulnerability of the periwinkle to the test chemical was determined using the Probit method of analysis for median lethal concentration LC_{50} at 10 days. In addition, the analysis of variance (ANOVA) in Statistical Package for Social Science (SPSS) statistical software in Version 21.0 was also used to test the mean statistical difference between the controls and treated groups at significance levels of $P < 0.05$. XY scattered graphs was used for the pictorial representation of experimental assessment.

RESULTS AND DISCUSSION

General observations under laboratory conditions: The snails generally remained on the surface of the sediment when food supply was sufficient. When dissolved oxygen in the overlying water was below 4.0 mg/L, the snails crawled up the test tanks. Under the present experimental conditions, the snails were activity and maintained the status throughout the 28 days.

Assessment of sediment toxicity: The results obtained from the acute toxicity of periwinkle snails exposed to different concentrations of ParaeForce® in spiked sediment are presented in Tables 2-3 and Figures 1-2. The results for the experimental bioassay showed that there was no death recorded in the control (Figure 2). Water quality parameters were within test method specified ranges. All the periwinkles snails were alive and healthy and showed movement when placed on a white clean paper or gently touched with platinum rod in control. The survival rates in the controls were 100%. However, mortality was observed in all concentrations of ParaeForce® in each test tank at day 10 of the bioassay. At the end of the bioassay, mortality rate in the ParaeForce® toxicity test concentrations of 0.125, 0.25, 0.5, 1.0 and 2.0 mg/kg were 27%, 43%, 53%, 77% and 87% (Figure 2). The 10-day LC_{50} (lethal concentration causing 50% deaths of the organism exposed to a chemical) was calculated using the mortality data at each of the different sediment-spiked concentration groups. Mortality results were generally consistent across the three replicate aquarium of each sediment-pesticide group. Each data point in the population-level dose response graph of Figure 2 represents the average percentage mortality data. A regression line was generated using these points, from which the LC_{50} estimate was then interpolated. Using the Probit analysis, the estimated average lethal concentration LC_{50} of ParaeForce® was 0.665 mg/kg (Finney, 1971). Based on the Organization of Economic Cooperation and Development (OECD) rating for contaminants in sediment matrix, pollutants can be classified using the ratings detailed in Table 2 (OECD, 2003). The environmentally tolerable concentrations, which is the safe limit was estimated at 10% of the LC_{50} value that is 0.0665 mg/kg. The Organization of Economic Cooperation and Development (OECD) rating for ParaeForce® was 1, which is super toxic (Table 2). The physico-chemical composition of the sediment showed an average pH value of 5. 90 ± 0.06 and total organic content (TOC) of 1.18 ± 0.12%, which indicates that the samples were slightly acidic.

Sub lethal exposure test for 28 days: Exposure of the test organisms to test toxicant (ParaeForce®) for sub lethal bioassay for 28 days to evaluate growth/inhibition rate are presented in Table 3 and Figure 2. The results of the present study indicate that the commercial formulation of paraquat (ParaeForce®) is toxic and has the potential to impair growth in periwinkles snails. At the end of the sub

Table 1. Laboratory conditions for sediment toxicity testing with *Tympanotonus fuscatus var radula.*

Parameter	Conditions
Substrate	sediment
Temperature	24 ± 1°C
Light	Wide-spectrum fluorescent lights
Illuminance	1000 lux
Photoperiod	12L: 12D
Culture chamber	52-L glass aquarium, 65 cm × 30 cm × 27 cm
Ratio of sediment to water	1:2 i.e 1000g of sediment to 2000ml of uncontaminated water
Overlying water	Brackish water
Overlying water quality checks	Temperature and dissolved oxygen (DO > 4.0 mg/L) daily; hardness, alkalinity, conductivity, pH, and total ammonia weekly
Renewal of overlying water	Recirculation, filtration, aeration, and routine supplement for water loss

Table 2. Sediment toxicity rating.

Rating	Designation	LC50 mg/g
1	Super toxic	<1.0
2	Extremely toxic	1.0-10
3	Very toxic	10-100
4	Slightly toxic	100-1000
5	Practically non-toxic	>1000

Table 3. Mean specific growth rate (weight) of periwinkles exposed to ParaeForce® in comparison with the control.

Concentration, mg/kg	Mean weight (g) ± SD	Specific growth rate (SGR) (g/d)	Mean percent growth rate relative to control (%)	Mean percentage growth rate inhibition efficiency (%)
Control	5.474 ± 0.04	0.1957	100	0
0.15	5.461 ± 0.06	0.1771	90	10
0.30	5.173 ± 0.03	0.1298	66	34
0.60	4.978 ± 0.02	0.0916	47	53

lethal bioassay exposure no death was observed for the 28-day period. However, there were changes in colour in the periwinkles with most of the species looking pale at test termination. Similarly, the morphology of the organisms was also affected, which result in very slow movement of the species when compared to the control. It is most likely the chemical have weakened effect on their locomotive potentials. In addition, the data revealed that there was also loss of body mass of the organisms especially in the highest sub lethal concentrations of 0.6 mg/kg (Table 2). This implied that the test herbicide induced poor feeding or low feed conversion ratio effects on the species. There was no significant alteration in the control treatments at levels of $P < 0.05$. In the sub lethal test for periwinkles, 10-53% reduction in weight was observed between the lowest and the highest sub lethal concentrations (Figure 2).

Inhibition efficiency (GIE): Paraeforce (e) a paraquat dichloride is a commonly used non-selective herbicide (Dogan and Can, 2011). According to the International Program for Chemical Safety Classification System of Pesticides by the World Health Organization (WHO), paraquat is classified as 'moderately hazardous' (WHO class II) (El-Sayed *et al.,* 2007). In this study ParaeForce® was super toxic Table 2. The herbicide industry claims that paraquat is safe if handled as instructed, yet paraquat poisoning remains a severe health problem globally and the degree of the severity depends on the exposure route and dose. In general, oral ingestion of paraquat is fatal as it is acutely toxic to humans. In ecotoxicology, 10-day LC_{50} for sediment toxicity is one of the most valuable parameters for assessing the toxic effects of pollutants. Herein, the10-day LC_{50} value (i.e. 0.665 mg/kg) was obtained from *Tympanotonus fuscatus var radula* exposed to paraquat (ParaeForce®), which suggests that this herbicide is highly toxic to gastropod snail. The observed snail mortality was both time and concentration dependent. The LC_{50} values of paraquat previously reported were 1.48 mg/L for *Mesopotamichthys sharpeyi* (Safahieh *et al.,* 2012), 1.41 mg/L for *Trichogaster trichopterus* (Banaee *et al.,* 2013), and 7.00 mg/L for *Oreochromis niloticus* (*O. niloticus*) (Ada *et al.,* 2012), respectively. No value has been reported for gastropod snail *Tympanotonus fuscatus var radula* prior to this study for comparison. The toxicity of a pesticide for an organism is affected by the strains of species, size, age, sex, temperature, water quality and formulation of the test chemicals (Pandey *et al.,* 2005, Nwani *et al.,* 2013). The sub lethal concentrations of paraquat used in our study (0.15, 0.30 and 0.60 mg/kg) are environmentally realistic because environmental concentrations of 0.01 µg/L and 0.14-8.70 µg/L of paraquat have been reported in Elechi Creek, Nigeria (Upadhi and Wokoma, 2012), and in some water bodies in Thailand (Tirado *et al.,* 2008), respectively. Our test concentrations was within the 0.1 µg/L safety limit established by the United State Environmental Protection Agency (Ladipo *et al.,* 2011), considering the repeated application of the pesticide in agricultural fields and other anthropogenic sources, the actual concentration in aquatic environments may exceed this limit.

The observed behavioural changes in *Tympanotonus fuscatus var radula* exposed to paraquat in the present study, which indicated slow movement, may be attributed to a neurotoxic effect of paraquat. Our results are consistent with previous reports on fish exposed to paraquat (Ladipo *et al.,* 2011; Ada *et al.,* 2012; Banaee *et al.,* 2013), and in other pesticides, such as carbosulfan (Altinok *et al.,* 2012), malathion (Ahmad, 2012), dichlorvos (Ahmad, 2012), and chlorpyrifos (Omoniyi *et al.,* 2013). Despite molluscs' species being the second largest group in Kingdom Animalia, they have not been considered in environmental risk assessment so far, mainly due to the lack of standardized protocols. Environmental change due to anthropogenic activities fundamentally affects individuals in complex networks of species. The emergent effects of environmental change on any one species therefore depend on its interactions with others (Sanford, 1999; Harley, 2011; Kroeker *et al.*, 2013). Molluscs are directly relevant to the human economy in Nigeria as they are use as food as good source of animal proteins.

Several efforts in Nigeria are being made to ensure that animal protein supply does not decrease irrespective of the increasing population size without commensurate increase in animal meat production. Snails have been reported to have high protein content and medicinal values, and as such, the demand for snail meat has increased over the years in domestic markets, thereby leaving behind lagging supply. Therefore, it is important to understand mollusc reactions to chemical compounds (OECD, 2010). The contamination of aquatic environments by herbicides has been characterized as a major ecological concern due to the indiscriminate use of these products in the control of aquatic plants and agricultural use. When applied, the chemicals tend to accumulate overtime and may pose certain effect on non-target aquatic species due to their toxicity. Aquatic environments represent more than two thirds of the Earth, and more than 90% of living animal species are invertebrates (Jha, 1998). Therefore, protecting aquatic invertebrates will play important roles in trophic chains by providing food for fishes, mammals, birds and man.

Tests developed with molluscs may be more amenable to extrapolation in risk assessment programs than those based on phyla less diverse and less numerically significant. In general, when herbicides contaminate the aquatic ecosystem, they can cause deleterious effects on the organisms system. Organisms that live in regions impacted by pesticide substances, whose breeding period coincides with the application period of the pesticides, can suffer serious risks of development and survival of their offspring.

Some of the side effects of pesticides exposures includes weight loss, decreased disease resistance, and increased susceptibility to predation, lack of interest in mating, destruction of chemoreceptor, imposex and defending territory (Ying and Williams, 2000). In the present study, the periwinkle species showed weight loss due poor feeding rate and low feed conversion ratio as a result of pesticide exposure. Exposure to sublethal concentrations revealed progressive decrease in body weight with mean decrease of 5.461 ± 0.06, 5.173 ± 0.03 and 4.978 ± 0.02 per cent respectively as opposed to 5.474 ± 0.04 per cent mean increase in the weight of control snail, at the end of the 28 day exposure. Statistical comparisons reveal that the percentage changes in body weight of test snail at all expo-

sure concentrations on every weekly measurement were significantly different from those of the control at $p < 0.0002$. Weight changes among the three exposure concentrations were also significantly different (Table 3). In the present study, the exposed snail maintained their feeding regime; hence the loss of weight cannot be correlated with starvation but may be due to an indirect effect of the toxicant on macromolecular syntheses which are secondary effects induced by physiological stress (Miliou *et al.,* 1998). Similar depression in growth, as measured by body length, was observed in Daphnia magna on exposure to endosulfan (Fernández-Casalderrey *et al.,* 1993).

Globally, the decline of gastropod population has been recorded as a consequence of the induction of the imposex effect in female molluscs by tributyltin (TBT) pollution (Chiavarini *et al.,* 2003). Tributyltin (TBT) pesticide has been recognised as one of the most toxic substances that have been deliberately introduced into natural waters (Goldberg, 1986; Terlizzi *et al.,* 2004). Also, most pesticides with heavy molecules that are not biodegradable are xenoestrogens or endocrine xenobiotics (Matthiessen *et al.,* 1995; Ogbomida and Ezemonye, 2013). It has been observed that the effects of pesticides do not occur only at the places that they are applied but also in places distant from their application and induce alterations in non-target organisms, altering the survival and equilibrium of the ecosystems, whether aquatic or terrestrial. Concentration of endocrine xenobiotics levels as low as 1 µg/L can induce pseudo-hermaphroditic condition in periwinkle snail species. Thus, much care must be taken when introducing these substances into the environment especially understanding their toxic consequences.

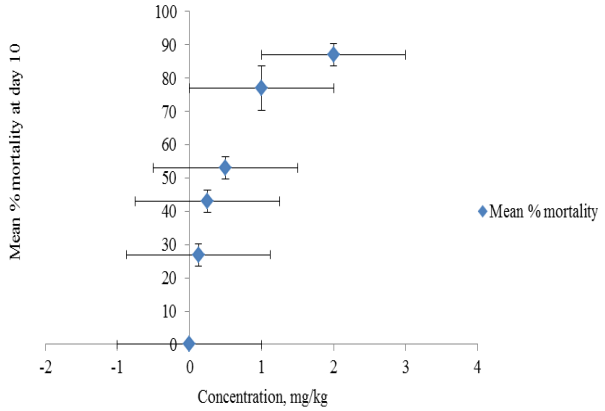

Figure 1. *Mean % mortality ± SE of periwinkle exposed to ParaeForce® at day 10.*

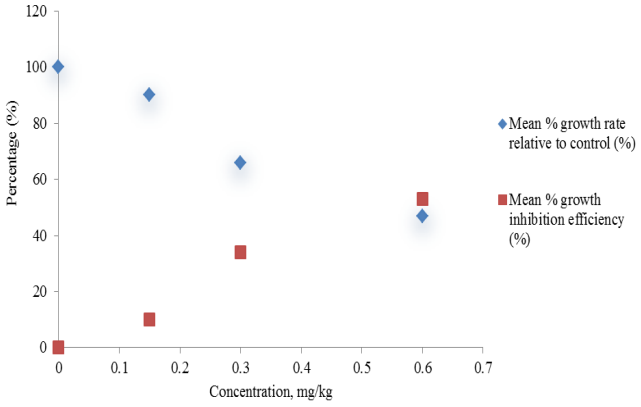

Figure 2. *Mean (%) growth rate relative of control (GR) in comparison with mean growth.*

Conclusions

In this assessment, exposure of periwinkle to ParaeForce® on short and long-termed bases caused detrimental effects on the species including mortality and loss of body mass. The test revealed that the increase in concentration of ParaeForce® in the sediment lead to an increase in the percentage mortality of the test organism periwinkle (*Tympanotonus fuscatus var radula*). It is therefore evident that the effect of acute toxicity of ParaeForce® is concentration related; the greater the concentration, the greater the effect. This implied that high concentration of these toxic substances in the environment would consequently pose great environmental and human health risks. National, regional and global awareness are needed to reduce the amount of chemicals entering the environment to reduce environmental and human exposure. In conclusion, the result of the study suggests that the toxic effects of ParaeForce® had adverse effects on non-target organism, thus, much care must be taken when introducing these substances into the environment. Further studies on the chronic effects of paraquat are still needed for a greater understanding of the environmental and health risks consequence. The use of paraquat at riverside and coastal areas should be strongly controlled and carefully monitored to avoid exposure to aquatic environments.

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